

## SOP for Standard Sequencing – Individual Reactions

If you have questions about how to organize a HT sequence request, please call John Elser at 301-846-6220 or Claudia Stewart at 301-846-5924

### Paper Work

Requests are made in the LMT-LIMS at:

<https://limsdev-abcc.ncifcrf.gov:8443/LIMS/custLogin.jsp>

Remember in the LMT-LIMS, you must first make at least one folder before you will be able to make a request.

Sample and request labels must be UNIX compatible text. This is Alphanumeric, dot, dash, underscore ONLY. No spaces please!

Use the COMMENTS field to provide additional information about your samples or to clarify anything that may seem confusing or unclear.

Once your request has been made, please go to the 'View Request' page and print the request you have just made and send along with the samples.

### Samples Preparation

LMT staff performs all the standard (tube) sequencing reactions

Samples must be submitted in 0.2ml PCR tubes (PE part # N801-0533) sealed with strip-caps (PE part # N801-0535). Please do not secure the lids with parafilm, this makes it more difficult to process the samples and labels can be worn off in the process of removing the parafilm. Our robotics system does not allow us to accept hinged lid tubes - so please understand that if we receive them we will have to return the samples to you. If you do not have the appropriate tube readily available, we can send some to you via courier or interoffice mail.

Template and Primer must be submitted separately.

Template Tube:           10 ul of 60-80 ng/ul plasmid DNA or  
                                  10 ul of 25-50 ng/ul PCR product

Primer Tube:             10 ul of 0.5 uM primer

We need one primer tube for each template tube, even if the same primer is to be used with multiple templates. To be very clear – please send a tube of primer and a tube of template for every reaction to be made.

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Primers, and their sequences, that can be provided by the LMT can be found at the following web location:

<http://web.ncifcrf.gov/atp/LMT/LMT-sequencing-Primers.pdf>

## Labeling-Tubes, Plates, Tube Holders

Tubes must be in a holder or rack designed for 0.2 ml tubes. We recommend the racks from Nunc product #251357. Holder must be labeled with Request Label from LMT-LIMS. We will return your holder if it is labeled with your Name, Building and Room.

Label the template tubes (not lids, we remove these to process the samples) with the requestor's initials, the tube or well designation from the LIMS request and T (for template). Example jd01-T, jd02-T for tube request; jdA01-T, jdA02-T for a plate request submitted by Jane Doe.

\*Label the primers tubes in a similar manner so as to match the template tube by substituting P for T. Example jd01-P, jd02-P, jdA01-P, jdA02-P would go with the above template samples.

Put your tubes in numerical order in the rack or holder with primer and template pairs together. This reduces the possibility of mixed up samples  
Please do not try write the entire sample name on the tube, we can't read it, and you will be frustrated trying. We need simple numbering so that we know at a glance that tubes are kept in the correct order.

## Shipping

Regulations require that we ask you to package and ship samples according to NIH guidelines.

For our Frederick customers to ship sequencing samples to Tollhouse Ave.:

- A. Safety requires a "Request for Shipment" form be filled out and sent to them prior to shipping any samples via the courier. A one time blanket form can be set-up for sending sequencing samples to Tollhouse Ave. which will streamline your paperwork. You can contact Sue Smith in Safety 301-846-5919 for details on this process.
- B. Call the courier by 4:30 pm the day before you plan on sending your samples. Let them know that you will have samples shipping to Tollhouse Ave. the next morning.
- C. Package your samples in an appropriate container and sufficient dry ice so that we receive them still frozen. Samples must be packed in a leak-proof container (Styrofoam box is OK). The lid must be secured with tape and labeled with both our address and phone number and your address and phone number
- D. Take your samples to the mail drop for your building by 9:30 am for pick-up.
- E. Samples that are not properly packaged (for example-in a zip-loc bag, in a mailing envelope, in an interdepartmental mailing envelope, taped together, in a 50 ml tube....) will not be accepted by the courier.

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For our Main Campus NIH customers to ship sequencing samples to Tollhouse Ave.:

- A. Call the courier by 9:30 am the day you plan on sending your samples. Give them your pick-up location and phone number as well as our address. Your phone number is very important, since couriers sometimes have difficulty accessing the NIH buildings due to the increased security. They may need to call you to let them into the building.
- B. Follow NIH guidelines for packaging and shipping.  
<http://www.nih.gov/od/ors/ds/shipping/index.html>
- C. Package your samples in an appropriate container and sufficient dry ice so that we receive them still frozen. Samples must be packed in a leak-proof container (Styrofoam box is OK). The lid must be secured with tape and labeled with both our address and phone number and your address and phone number.
- D. Have your package ready by 11:00 am.
- E. Samples that are not properly packaged (for example-in a zip-loc bag, in a mailing envelope, in an interdepartmental mailing envelope, taped together, in a 50 ml tube....) can not be accepted by the courier.

Our shipping address is:  
LMT Sequencing Lab  
915 Tollhouse Ave.  
Suite 211  
Frederick, MD 21701

Our phone number is:  
301-846-5676  
and Fax:  
301-846-6100

## LMT Staff Questions

If there are any problems or questions about your samples, or if there are any questions about your request, you will be notified by the LMT staff. Your request will show a status of 'Received and Waiting' until issues are resolved and this may increase your turnaround time. In the rare instance that a problem can not be resolved by e-mail or phone, your samples will be returned to you.

## Special Handling

If you need to submit samples in any thing other than this standard format, you must get pre-approval from the LMT staff. This will be treated as a custom request and must include specific details along with your samples.

## **Related Web Sites**

LMT web site <http://web.ncifcrf.gov/rtp/lmt/>

NCI web site <http://web.ncifcrf.gov>

LMT LIMS <https://lmsdev.ncifcrf.gov:8443/LIMS/custLogin.jsp>